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Processing Effects on Selected Antioxidant Activities and Metabolizing Enzyme Inhibition of *M. Koneigii* (Curry Leaves) Extracts

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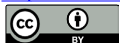

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Abstract

Curry leaves, scientifically termed *Murraya koenigii*, are renowned in South Asian cuisine for their flavor enhancement and potential health benefits, including antioxidative, anti-inflammatory, and antidiabetic properties. This study aimed to evaluate the impact of thermal processing methods on curry leaves by analysing Total Phenolic Content (TPC), Total Flavonoid Content (TFC), antioxidant activity, and metabolizing enzyme inhibition. Fresh curry leaves were subjected to thermal treatments: Oven-dried at 60˚C and Air-dried at 25˚C for 2 weeks. Extracts were prepared using Ethanol and water solvents. Results indicated that Air-dried leaves exhibited significantly higher TPC (5132.65 mg GAE/100 g) and TFC (243.13 mg CE/100 g) compared to Fresh and Oven-dried leaves. Antioxidant assays show that oven-dried curry leaves at 60˚C displayed higher results in NORS, FRAP, and TEAC assays compared to Fresh and Air-dried leaves. Ethanol extracts showed better extraction of bioactive compounds than aqueous extracts. Moreover, Lipase inhibition activity was notably high, indicating potential health benefits. This study provides valuable insights into the effects of processing methods on curry leaf extracts, emphasizing the importance of solvent selection for optimal extraction of bioactive compounds.

Keywords

Murraya Koenigii, Curry leaves, Antioxidants, Phytochemicals, Metabolizing Enzymes

1. Introduction

Plants and their components have been utilized in a variety of ways: used as

food, fodder, medicine, wood, etc. According to the World Health Organization (WHO), almost one-fifth of the world's population uses plant-based medicines (Phytomedicines) for primary health care [1]. Phytochemicals are non-nutritive substances produced by plants that have therapeutic or disease-prevention properties. Plants create these substances to protect themselves, but a recent study indicates that several phytochemicals may also protect against diseases that affect humans [2].

Some diseases that occur in humans also tend to impact biological functioning. One such example is the severe oxidative damage to biological macromolecules such as DNA, proteins, lipids, etc., caused by reactive oxygen species (ROS) and free transition metal ions. As a result, such biological changes might also lead to the pathophysiology of diseases caused by oxidative stress [3] [4].

Antioxidants are crucial in biological systems [5]. Due to their ability to suppress the generation of reactive oxygen species by decreasing hydroperoxides (ROO•) and Hydrogen peroxide and scavenging free radicals, among other things [6]. Natural substances originating from plants, especially those that can quench singlet and triplet oxygen, may also act as enzyme inhibitors, peroxide decomposers, and synergists [7] [8].

Murraya koenigii (family: Rutaceae), also known as the curry leaf plant native to India, Sri Lanka, and various other South Asian nations [9]. *Murraya koenigii* leaves are widely recognized plant-based spices used in tiny doses for their distinctive aroma due to the concentration of volatile oil, and for their potential for aiding digestion [10]. Plant phenolics, including tannins, flavonoids, and phenolics, are renowned antioxidant compounds [11]. Previous empirical research findings [12] indicate that curry leaves at an advanced stage of maturity or full development exhibit more accumulation of antioxidants showing more scavenging activity relative to immature or tender leaves. Aromatic bioactive compounds in *M. koenigii* leaves preserve flavor and other properties even after drying [13]-[15].

Drying is a preliminary food preservation procedure that reduces the amount of water in a product to a level that prevents microbial growth, hence improving the shelf life of perishable plant-based raw materials [16]. Moisture content is an important component in determining microbiological safety between storage and consumption. As a result, decreasing water activity is a key step in preserving the nutritional value of herbs [17]. In addition, drying also reduces the weight of the product, thereby reducing the amount of packaging needed and facilitating easier transport and storage [18].

The Western diet is rich in simple carbohydrates, increasing the risk of cardiovascular disease, diabetes, and cancer. α-Amylase and α-glucosidase are important enzymes required in the body for starch breakdown and intestinal absorption [19]. α-Amylase degrades long-chain carbohydrates, whereas α-glucosidase degrades starch and disaccharides to produce glucose [20]. Pancreatic lipase, also known as pancreatic triacylglyceride lipase, is a crucial functional

enzyme in the conversion of triacylglycerides to monoacylglycerides and fatty acids [21].

Scientific studies [22] show that alcoholic extraction of curry leaves is more effective than water extraction at isolating medicinally beneficial components such as cardiac glycosides, antioxidants, and essential oils. Moreover, other parts of *M. koenigii*, such as roots, bark, and fruit, have also been shown to support various biological activities [23]. This research focuses on the concentration of plant-based antioxidants in curry leaves that may neutralize free radicals and have protective properties against cardiovascular diseases using Aqueous and Ethanol solvent extraction methods.

2. Materials and Methods

2.1. Instruments Used

Hot air oven (Isotemp Oven, Fisher Marietta OH, model 6925), **Freeze dryer** (VirTis Genesis 35L SpScientific, Warminster, PA), **Electronic magnetic Stirrers** (MTX-15, 2 mag USA), **Centrifuge** (Thermo Scientific, 2011, sorvall legend XTR), **Rotary evaporator** (Buchi Rotavapor R-215, USA), **Waring Blender** (Model no. 31BL92, New Hartford, Connecticut, US), **Microplate reader** (Biotek, Instrument Inc, Synergy MTX, Winooski, VT).

2.2. Sample Preparation

To perform the sample preparation, fresh curry leaves were first purchased from the local market. The leaves were then separated from the rachis, washed, and cleaned. Subsequently, the processed leaves were subjected to drying.

2.2.1. Freeze Drying

In this method, the washed and cleaned curry leaves were first placed in the freezer at −80˚C for 2 hours. They were evenly spread in a single layer of leaves on trays and then dried in the freeze dryer (VirTis Genesis 35L Sp Scientific, Warminster, PA). Dried curry leaves were then converted into a powder form using a Waring blender (Model no. 31BL92, New Hartford, Connecticut, US) and stored for future use.

2.2.2. Oven-Drying

For the oven-drying method, curry leaves were spread on trays followed by drying in the oven (Isotemp Oven, Fisher Marietta OH, model 6925) at a selected temperature (60˚C) for 3 hours. Dried curry leaves were then converted into powdered form using a Waring blender (Model no. 31BL92, New Hartford, Connecticut, US) and stored for future use.

2.2.3. Air-Drying

Shade-drying procedures that have been employed in the past have been utilized. Leaves were separated from the stalks and then washed with water. Leaves were then scattered on a tray in a single layer and air-dried for 14 days at an ambient

room temperature of 25˚C. After collecting the dried leaves, they were ground into a fine powder in a Waring blender (Model no. 31BL92, New Hartford, Connecticut, US), weighed, and packaged into a polyethylene container for future use.

2.2.4. Moisture Content

For the moisture content analysis, the standard protocol by AOAC (Association of Official Analytical Chemists) (AOAC, 2005; method 930.15) was followed [24].

2.3. Solvent Extraction

Aqueous and ethanol solvent extraction were prepared following the method developed by [25] with slight modifications. Five grams of dried curry leaves were agitated for two hours in 50 ml of 80% ethanol and deionized water. The supernatant was recovered from agitated leaves by centrifuging the samples at 3000 g for 20 minutes. The supernatant was then evaporated to dryness at 50˚C by a rotary evaporator (Buchi Rotavapor R-215, USA). For further examination, samples were resuspended in 10 ml of 80% ethanol and water before being stored at −80˚C.

3. Chemical Analysis

3.1. Total Phenolic Content

The total phenolic content of curry leaf extracts was determined using a modified Folin-Ciocalteu (FC) colorimetric technique [26].

3.2. Total Flavonoid Content

For the determination of the Total Flavonoid Content, Catechin was used as the standard for calculating the flavonoid content in the sample assays, with slight modifications [26].

3.3. Antioxidant Activity

3.3.1. DPPH 2,2-Diphenyl-1-Picrylhydrazyl (DPPH)

A standard protocol [27] was followed with slight modifications for the determination of DPPH 2,2-diphenyl-1-picrylhydrazyl Radical Scavenging Ability.

3.3.2. Ferric Reducing Antioxidant Potential (FRAP)

For the determination of Ferric Reducing Antioxidant Potential, a standard protocol was developed by [28] with slight modifications.

3.3.3. Trolox Equivalent Antioxidant Capacity (TEAC)

The determination of Trolox Equivalent Antioxidant Capacity was followed by using a standard protocol developed by [29] with slight modifications.

3.3.4. Nitric Oxide Radical Scavenging (NORS) Activity

The determination of Nitric Oxide Radical Scavenging Activity assay was fol-

lowed by using a method developed by [30] with slight modifications.

4. Enzymatic Analysis

4.1. Alpha Amylase Inhibition Activity

The inhibition of Alpha-amylase in extracts was determined using the methodology of [31] with minor modifications.

4.2. Alpha Glucosidase Activity

The inhibition of α-glucosidase in extracts was followed by using the modified approach suggested by [31].

4.3. Lipase Activity

Lipase inhibition of extracts was measured using DNPB as a substrate by [32].

5. Statistical Analysis

All experiments were carried out in triplicates, and data were recorded as means ± standard deviation. For the chemical analysis of curry leaves extracts, factorial design (3*2) was followed with 3 treatments (Fresh, Oven-dried at 60˚C, and Air-dried), and 2 solvents (Aqueous and Ethanol) for extraction of bioactive compounds. For the enzymatic analysis of curry leaves extracts, factorial design (3*2) was followed with 3 treatments (Fresh, Oven-dried at 60˚C, and Air-dried), and 2 solvents (Aqueous and Ethanol). SAS 9.2 was used to conduct ANOVA (Analysis of variance). Tukey's studentized range test was used to evaluate significant differences between means, and ($p \leq 0.05$) considered as significant. (**Figure 1**)

6. Results and Discussion

6.1. Moisture Content

In their fresh state, the moisture content in curry leaves is 54.27% of total dry weight. The thermal processing treatments influenced the moisture content. When the samples were oven-dried at 60˚C, the moisture content was lowered to 34.12%. In contrast, when the samples were air-dried, the moisture content was lowest at 15.39%.

6.2. Total Phenolic and Flavonoid Content

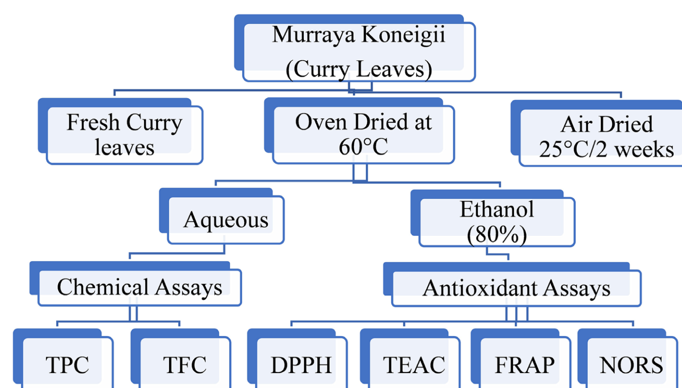
This section presents the total content of polyphenols and flavonoids in fresh curry leaf extracts (F), oven-dried at 60˚C curry leaves extracts (OD) and air-dried curry leaves extracts (AD) when using Aqueous (A) and Ethanol (E) solvents respectively. The results are summarized in **Table 1**.

There were significant ($p \leq 0.05$) differences between the solvent type and thermal processing methods for the phenolics. The total phenolic content was higher in Ethanol solvent extracts when compared to Aqueous solvent extracts, where

TPC for Ethanol extracts was about 32 folds higher than Aqueous for Fresh curry leaves extracts. Additionally, it was 10 folds and 65 folds higher for OD and AD. Phenolic compounds have been shown to contribute to the antioxidant properties of plant substances [33].

Previous research indicates that the increased phenolic content found in alcohol: water (1:1) extracts of curry leaves could potentially account for the heightened antioxidant effects observed in comparison to other extracts [34]. In similar studies, it has been observed that the increased presence of phenolic compounds in plant extracts is associated with higher antioxidant effectiveness [35].

Likewise, a similar trend of significant ($p \le 0.05$) differences in flavonoid content was also seen for the different solvent types and thermal processing methods. The flavonoid content in ethanol extracts was about 66% higher than Aqueous for Fresh curry leaves extracts. Similarly, it was about 273% and 30% higher for OD and AD. These results show more extractability when using Ethanol solvents than water solvents.



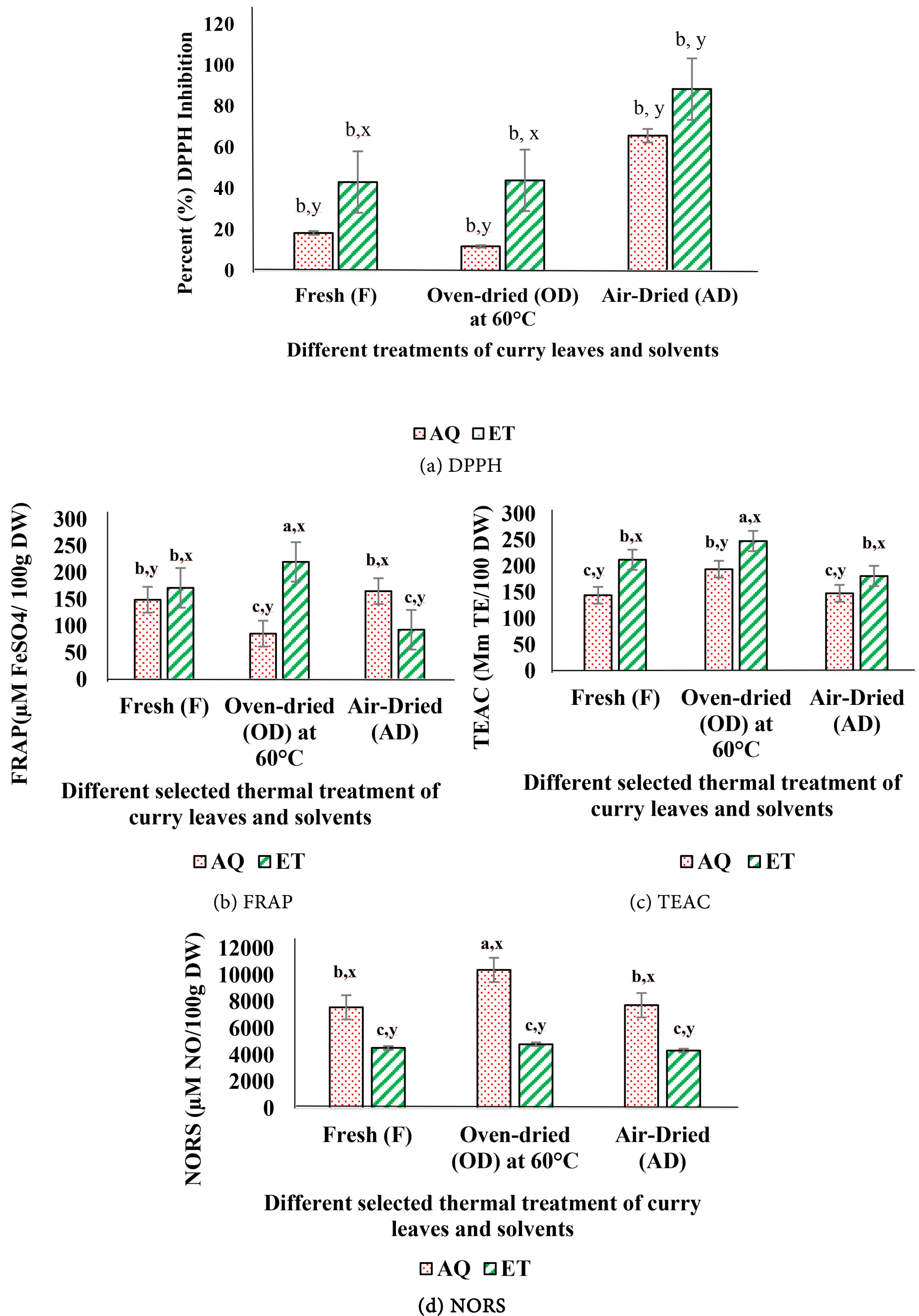
(TPC—Total Phenolic Content, TFC—Total Flavonoid Content, DPPH— 2,2-diphenyl-1-picrylhydrazyl, FRAP—Ferric Reducing Antioxidant Potential, TEAC—Trolox Equivalent Antioxidant Capacity, NORS—Nitric Oxide Radical Scavenging Ability)

Figure 1. Experimental design (3*2 Factorial) for chemical analysis.

Table 1. Total Phenolic content and Total Flavonoid Content of aqueous and extracts of selected thermally treated curry leaves.

Chemical Analysis	*Solvent*	*Fresh (F)*	*Oven-dried at 60˚C (OD)*	*Air-dried (AD)*
TPC (mg GAE/100 g DW)	*Aqueous (A)*	$113.95 \pm 41.26^{b,x}$	$322.66 \pm 109.15^{a,x}$	$79.09 \pm 27.10^{c,y}$
	Ethanol (E)	$3605.91 \pm 577.02^{b,x}$	$3288.13 \pm 390.12^{b,x}$	$5132.65 \pm 915.65^{a,x}$
TFC (mg CE/100 g DW)	*Aqueous (A)*	$56.42 \pm 9.91^{b,y}$	$51.87 \pm 9.19^{b,y}$	$186.87 \pm 30.02^{a,y}$
	Ethanol (E)	$93.70 \pm 10.08^{c,y}$	$193.27 \pm 49.99^{b,x}$	$243.13 \pm 80.47^{a,y}$

TPC—Total Phenolic Content, TFC—Total Flavonoid content, GAE-Gallic Acid Standard, CE—Catechin Equivalent Standard, DW—Dry Weight. Significant differences ($p \le 0.05$) among thermal treatments within curry leaf extracts types are indicated using the letters 'abc' and solvent 'xy'



(a) DPPH

(b) FRAP

(c) TEAC

(d) NORS

(a) DPPH—2,2-Diphenyl-1-Picrylhydrazyl (b) FRAP-Ferric Reducing Antioxidant Capacity, (c) TEAC— Trolox Equivalent Antioxidant Capacity, (d) NORS—Nitric oxide radical scavenging, DW—Dry Weight. Treatments-Fresh (F), Oven-dried at 60˚C, Air-Dried (AD), Solvents: AQ-Aqueous ET-Ethanol 80% Bars with superscripts: a,b,c-(Thermal Processing), x,y-solvents are significantly different ($p \leq 0.05$).

Figure 2. Antioxidant activities of selected thermal processing treatment of curry leaves extracts.

6.3. Antioxidant Activity of Curry Leaves Extracts

6.3.1. (2,2-DIPHENYL-1-PICRYLHYDRAZYL (DDPH) Radical Scavenging of Curry Leaves Extracts

Compounds capable of donating hydrogen or electrons to DPPH, a nitrogen-centered free radical, are recognized as antioxidants, acting as radical scavengers. The extent of the reduction in the violet color of DPPH reflects the antioxidant's ability to scavenge radicals [36]. DPPH

(2,2-DIPHENYL-1-PICRYLHYDRAZYL) radical scavenging activity of curry leaves extracts ranged from 11.84% to 88.41% inhibition, which represents antioxidant activity for different drying treatments as shown in **Figure 2**. Furthermore, the scavenging activity was higher when using Ethanol extracts compared to Aqueous extracts. The observed values for Ethanol solvents were at least double, 4 times, and 1.5 times the value obtained for Aqueous solvents in fresh, OD, and AD respectively. The range of observed inhibition percentages were slightly lower when compared to previous studies [34]. This indicates a higher free radical scavenging capacity of curry leaves, which is desirable for assessing the antioxidant activity and thus providing better pharmaceutical application.

6.3.2. Ferric Reducing Antioxidant Power (FRAP) of Curry Leaves Extracts

The FRAP results are also presented in **Figure 2(b)**. The ability to convert $Fe3+$ to $Fe2+$ through reduction serves as an indirect indicator of the antioxidant potential of an extract or compound [37]. While the FRAP activity in Ethanol solvent for Fresh and OD at 60˚C was higher than in Aqueous solvent, the trend was reversed for AD. Specifically, the observed FRAP values were about 15% and 156% higher in Ethanol for fresh and OD respectively, when compared to Aqueous. On the contrary, the FRAP values were about 77.5% higher in Aqueous for AD compared to Ethanol extracts. Previous research has shown that ethanol extracts of curry leaves exhibit the most significant reducing power among all tested samples when compared to alternative solvents [38]. This highlights the variability in FRAP values between (E) and (A) extracts under different drying conditions, indicating that the choice of solvent and sample preparation method can significantly impact the antioxidant activity measured by the FRAP assay.

6.3.3. Trolox Equivalent Antioxidant Capacity (TEAC) of Curry Leaves Extracts

TEAC values significantly ($p \leq 0.05$) increased by approximately 46.9% in Ethanol extracts of fresh curry leaves when compared to Aqueous extracts. In curry leaves extracts for oven-dried at 60˚C, TEAC follows a similar trend of higher values for Ethanol when compared to Aqueous with an increase of approximately 27.7%. Likewise, for the air-dried curry leaves extracts, TEAC values were significantly ($p \leq 0.05$) higher (22.3%) in Ethanol extracts when compared to Aqueous extracts. Since a higher TEAC value indicates greater antioxidant activity, implying that the substance has a stronger ability to neutralize free radicals and oxidative stress. While the TEAC was higher for Fresh curry leaves but

the difference was also significant when using either OD at 60˚C or AD.

6.3.4. Nitric Oxide Radical Scavenging (NORS) of Curry Leaves Extracts

Excessively elevated concentrations of nitric oxide (NO) have been associated with persistent inflammation and connected to the cause and development of several long-term chronic diseases [39]. Results of the NORS assay are also shown in **Figure 2(d)**. From the observed values in Fresh, OD or AD curry leaves extracts, NORS values were significantly ($p \le 0.05$) higher for Aqueous solvent when compared to Ethanol solvent. NORS value was 1.67-foldhigher in the fresh curry leaf extracts when compared to Aqueous Fresh curry leaf extracts. However, a 2.16-foldhigher value was observed in Aqueous extracts of OD at 60˚C when compared to the Ethanol extracts. Similarly, there was a 1.78-fold increase in the AD Aqueous extract compared to Ethanol extracts. These results indicate that higher NORS value indicate greater effectiveness in neutralizing nitric oxide radicals, reflecting stronger antioxidant and anti-inflammatory properties. The observed NORS values in **Figure 2(d)** indicate a highest potential with OD at 60˚C. Previous studies have shown that extracts of *M. koenigii* demonstrated inhibitory effects on nitric oxide production and nitric oxide radicals generated from sodium nitroprusside at physiological pH. Notably, these extracts, across concentrations ranging from 40 to 400 µg/ml, exhibited significant nitric oxide radical scavenging activity, with the scavenging activity percentage escalating with higher concentrations [38].

6.4. Carbohydrate and Lipid Metabolizing Enzyme Inhibition by Curry Leaves Extracts

The inhibition of selected metabolizing enzymes by Aqueous and Ethanol curry leaves extracts is shown in **Table 2**. Specifically, the percentage values of *α*-glucosidase, *α*-Amylase, and Lipase inhibitory activity for the extracts when using Fresh, OD at 60˚C, and AD treatments were obtained.

Table 2. Inhibition of selected metabolic enzymes by aqueous and ethanol curry leaves extracts.

Metabolizing enzymes	*Solvent*	*Fresh* (*F*)	*Oven-dried at 60˚C* (*OD*)	*Air-dried* (*AD*)
***α-glucosidase inhibition* (%)**	*Aqueous* (*A*)	$80.15 \pm 4.98^{a,x}$	$70.102 \pm 4.99^{b,x}$	$41.162 \pm 4.60^{c,y}$
	Ethanol (*E*)	$64.91 \pm 3.47^{a,y}$	$43.269 \pm 1.54^{c,y}$	$57.941 \pm 7.93^{b,x}$
***α-Amylase inhibition* (%)**	*Aqueous* (*A*)	$25.28 \pm 7.06^{b,y}$	$82.22 \pm 0.48^{a,x}$	$77.931 \pm 0.60^{a,y}$
	Ethanol (*E*)	$74.46 \pm 2.79^{b,x}$	$75.167 \pm 2.24^{b,y}$	$84.882 \pm 1.36^{a,x}$
***Lipase inhibition* (%)**	*Aqueous* (*A*)	$20.53 \pm 1.39^{a,y}$	$13.755 \pm 1.77^{b,y}$	$16.058 \pm 1.11^{b,y}$
	Ethanol (*E*)	$46.81 \pm 6.93^{b,x}$	$70.346 \pm 0.52^{a,x}$	$78.03 \pm 34.19^{a,x}$

*F-Fresh, Oven-dried at 60˚C (OD), Air-dried (AD) A-Aqueous E-Ethanol 80%.

Significant differences ($p < 0.05$) of thermal treatment in curry leaves extracts shown in rows with superscripts: abc, Significant differences ($p < 0.05$) of solvents shown in columns. The data was averaged in triplicates ($n = 3$).

6.4.1. *α*-Glucosidase Inhibitionby Curry Leaves Extracts

Natural substances remain the most easily accessible source of glucosidase inhibitors [40]. Recent findings [41] show that Polyphenolic compounds slow down the absorption of glucose by inhibiting the activity of the enzymes *α*-amylase and *α*-glucosidase, which play a crucial role in the digestion of carbohydrates. The concentration of the sample used was 0.025µg/ml. The inhibition of *α*-glucosidase enzyme by the curry leaf extracts ranged from 43.26% to 64.91% for the Ethanol curry leaves extracts whereas for Aqueous solvent of curry leaf extracts ranged from 41.16% to 80.15%. For *α*-glucosidase inhibition, Fresh curry leaf extracts showed higher inhibition compared to OD at 60˚C and AD treatments. Among the solvents, Aqueous extract (A) showed lower inhibition than Ethanol extracts (E).

6.4.2. *α*-Amylase Inhibition by Curry Leaves Extracts

For *α*-amylase inhibition, OD at 60˚C and AD treatments generally show higher inhibition compared to Fresh. Within the Aqueous extract, the *α*-Amylase % inhibition significantly increased from Fresh (25.28%) to OD at 60˚C (82.22%) but was lower in AD (77.93%).For Ethanol extracts, there were no significant differences observed among the thermal treatments, as indicated by the similar values for Fresh (74.46%), OD at 60˚C (75.16%), and AD (84.88%).

6.4.3. Lipase Inhibition by Curry Leaves Extracts

As seen in **Table 2** for lipase inhibition, curry leaves extract for OD at 60˚C showed lower inhibition compared to fresh leaves and AD treatments. Furthermore, the inhibition of Lipase enzyme was better when the Aqueous solvent was used. Specifically, the lipase inhibition by the curry leaf extracts ranged from 13.76% in OD at 60˚C to 16.1% in AD and 20.53% in fresh leaves. For the Ethanol solvent, there were significant differences when comparing thermal treatments. The lipase inhibition significantly increased in Fresh (46.81%) to OD at 60˚C (70.34%), and a similar significant increase was observed when comparing Fresh (46.81%) to AD (78.03%).

7. Discussions

The presence of more phenolic compounds in the extracts of plants is linked to greater antioxidant activity [34]. The elevated polyphenolic content in the water-ethanol extract of curry leaves could suggest its antioxidant properties through the combined effect of all its components, rather than being solely attributed to a single constituent within the extract [42]. Curry leaves contain carbazole alkaloids such as murrayanine, mahanimbine, girinimbine, mukonine, and murrayafoline-A [34], four additional alkaloids namely euchrestine B, bismurrayafoline E, mahanine, and mahanimbicine [43] which have been identified as potential bioactive compounds. Plant extracts have the potential to serve as sources of electrons and engage with free radicals, transforming them into more stable substances and concluding radical chain reactions [35]. Comparable research has indicated that natural antioxidants play a role in preventing free rad-

ical reactions and demonstrate diminished efficacy [44]. The reduced capacity might be attributed to their ability to donate hydrogen [45]. Previous studies indicated that alcohol-water extract of curry leaves exhibited a total phenolic content of 168 mg/g [46]. In their research, they discovered that the alcohol and water extracts displayed the highest reductive potential, assessed through ferric ion reduction, while the hexane extract exhibited the lowest reductive potential.

8. Conclusion

The existing pre-clinical information in the literature suggests the impressive pharmacological potential of *M. koenigii*, rendering it a viable option for enhancing the prevention of certain diseases. The solvent extract from the plant holds significant medicinal potential. However, a comprehensive examination employing animal models and clinical trials is necessary to delve into the precise molecular mechanisms of action and assess its effectiveness against cytotoxicity assays. This quest is aimed at discovering promising compounds from natural sources that will aid in the reduction of chronic diseases.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

[1] Ahmad, I., Aqil, F. and Owais, M. (2006) Modern phytomedicine: Turning Medicinal Plants into Drugs. John Wiley & Sons. https://doi.org/10.1002/9783527609987

[2] Tomar, R.S., Banerjee, S. and Kaushik, S. (2017) Assessment of Antioxidant Activity of Leaves of *Murraya koenigii* Extracts and Its Comparative Efficacy Analysis in Different Solvents. *Journal of Pharmaceutical Sciences and Research*, **9**, 288-291.

[3] Dröge, W. (2002) Free Radicals in the Physiological Control of Cell Function. *Physiological Reviews*, **82**, 47-95. https://doi.org/10.1152/physrev.00018.2001

[4] Hippeli, S. and Elstner, E.F. (1999) Transition Metal Ion-Catalyzed Oxygen Activation during Pathogenic Processes. *FEBS Letters*, **443**, 1-7. https://doi.org/10.1016/s0014-5793(98)01665-2

[5] Cheeseman, K.H. and Slater, T.F. (1993) An Introduction to Free Radical Biochemistry. *British Medical Bulletin*, **49**, 481-493. https://doi.org/10.1093/oxfordjournals.bmb.a072625

[6] Gilgun-Sherki, Y., Melamed, E. and Offen, D. (2001) Oxidative Stress Induced-Neurodegenerative Diseases: The Need for Antioxidants That Penetrate the Blood Brain Barrier. *Neuropharmacology*, **40**, 959-975. https://doi.org/10.1016/s0028-3908(01)00019-3

[7] Larson, R.A. (1988) The Antioxidants of Higher Plants. *Phytochemistry*, **27**, 969-978. https://doi.org/10.1016/0031-9422(88)80254-1

[8] Mitrovic, A. and Bogdanovic, J. (2009) Effect of Gibberellic Acid on Total Antioxidant Activity during *Chenopodium rubrum* L. Ontogenesis *in Vitro*. *Archives of Biological Sciences*, **61**, 49-55. https://doi.org/10.2298/abs0901049m

[9] Khanum, F., Anilakumar, K.R., Sudarshana Krishna, K.R., Viswanathan, K.R. and

Santhanam, K. (2000) Anticarcinogenic Effects of Curry Leaves in Dimethylhydrazine-Treated Rats. *Plant Foods for Human Nutrition*, **55**, 347-355. https://doi.org/10.1023/a:1008148531495

[10] Jain, V., Momin, M. and Laddha, K. (2012) *Murraya koenigii*: An Updated Review. *International Journal of Ayurvedic and Herbal Medicine*, **2**, 607-627.

[11] Farasat, M., Khavari-Nejad, R., Nabavi, S.M.B. and Namjooyan, F. (2013) Antioxidant Properties of Two Edible Green Seaweeds from Northern Coasts of the Persian Gulf. *Jundishapur Journal of Natural Pharmaceutical Products*, **8**, 47-52. https://doi.org/10.5812/jjnpp.7736

[12] Abeysinghe, D.T., Alwis, D.D.D.H., Kumara, K.A.H. and Chandrika, U.G. (2021) Nutritive Importance and Therapeutics Uses of Three Different Varieties (*Murraya koenigii*, *Micromelum minutum*, and *Clausena indica*) of Curry Leaves: An Updated Review. *Evidence-Based Complementary and Alternative Medicine*, **2021**, Article ID: 5523252. https://doi.org/10.1155/2021/5523252

[13] Amna, U., Halimatussakdiah Wahyuningsih, P., Saidi, N. and Nasution, R. (2019) Evaluation of Cytotoxic Activity from Temurui (*Murraya koenigii* [Linn.] Spreng) Leaf Extracts against Hela Cell Line Using MTT Assay. *Journal of Advanced Pharmaceutical Technology & Research*, **10**, 5-55. https://doi.org/10.4103/japtr.japtr_373_18

[14] Husna, F., Suyatna, F., Arozal, W. and Poerwaningsih, E. (2018) Anti-Diabetic Potential of *Murraya koenigii* (L.) and Its Antioxidant Capacity in Nicotinamide-Streptozotocin Induced Diabetic Rats. *Drug Research*, **68**, 631-636. https://doi.org/10.1055/a-0620-8210

[15] Das, R., Bhattacharya, K., Sarkar, S., Samanta, S.K., Pal, B.C. and Mandal, C. (2013) RETRACTED ARTICLE: Mahanine Synergistically Enhances Cytotoxicity of 5-Fluorouracil through Ros-Mediated Activation of PTEN and P53/p73 in Colon Carcinoma. *Apoptosis*, **19**, 149-164. https://doi.org/10.1007/s10495-013-0907-6

[16] Mahayothee, B., Thamsala, T., Khuwijitjaru, P. and Janjai, S. (2020) Effect of Drying Temperature and Drying Method on Drying Rate and Bioactive Compounds in Cassumunar Ginger (*Zingiber montanum*). *Journal of Applied Research on Medicinal and Aromatic Plants*, **18**, Article ID: 100262. https://doi.org/10.1016/j.jarmap.2020.100262

[17] Kulshrestha, R., Gupta, C., Shukla, G., Kundu, M., Bhatnagar, S. and Katiyar, C. (2008) The Effect of Water Activity and Storage Temperature on the Growth of *Aspergillus flavus*in Medicinal Herbs. *Planta Medica*, **74**, 1308-1315. https://doi.org/10.1055/s-2008-1074561

[18] Kaur, R., Kaur, K. and Ahluwalia, P. (2020) Effect of Drying Temperatures and Storage on Chemical and Bioactive Attributes of Dried Tomato and Sweet Pepper. *LWT*, **117**, Article ID: 108604. https://doi.org/10.1016/j.lwt.2019.108604

[19] Kashtoh, H. and Baek, K. (2023) New Insights into the Latest Advancement in *α*-Amylase Inhibitors of Plant Origin with Anti-Diabetic Effects. *Plants*, **12**, Article 2944. https://doi.org/10.3390/plants12162944

[20] Ojah, E.O., Moronkola, D.O. and Mukaram, A. (2020) *α*-Amylase and *α*-Glucosidase Antidiabetic Potential of Ten Essential Oils from Calophyllum Inophyllum Linn. *Iberoamerican Journal of Medicine*, **2**, 253-260. https://doi.org/10.53986/ibjm.2020.0046

[21] Bajes, H.R., Almasri, I. and Bustanji, Y. (2020) Plant Products and Their Inhibitory Activity against Pancreatic Lipase. *Revista Brasileira de Farmacognosia*, **30**, 321-330. https://doi.org/10.1007/s43450-020-00055-z

[22] Sharma, R. and Kumar, U. (2019) Exploration and Phytochemical Estimation of *Murraya koenigii* Leaves for Pharmaceutical Applications. *Asian Journal of Pharmaceutical Research*, **9**, 159-168. https://doi.org/10.5958/2231-5691.2019.00025.x

[23] Balakrishnan, R., Vijayraja, D., Jo, S.H., Ganesan, P., Su-Kim, I. and Choi, D.K. (2020) Medicinal Profile, Phytochemistry, and Pharmacological Activities of *Murraya koenigii* and Its Primary Bioactive Compounds. *Antioxidants*, **9**, Article 101. https://doi.org/10.3390/antiox9020101

[24] Ahn, J.Y., Kil, D.Y., Kong, C. and Kim, B.G. (2014) Comparison of Oven-Drying Methods for Determination of Moisture Content in Feed Ingredients. *Asian-Australasian Journal of Animal Sciences*, **27**, 1615-1622. https://doi.org/10.5713/ajas.2014.14305

[25] Amazu, L., Azikiwe, C., Njoku, C., Osuala, F., Nwosu, P., Ajugwo, A., *et al.* (2010) Antiinflammatory Activity of the Methanolic Extract of the Seeds of *Carica papaya* in Experimental Animals. *Asian Pacific Journal of Tropical Medicine*, **3**, 884-886. https://doi.org/10.1016/s1995-7645(10)60212-x

[26] Gajula, D., Verghese, M., Boateng, J., Walker, L.T., Shackelfor, L., Mentreddy, S.R., et al. (2009) Determination of Total Phenolics, Flavonoids and Antioxidant and Chemopreventive Potential of Basil (*Ocimum basilicum* L. and *Ocimum tenuiflorum* L.). *International Journal of Cancer Research*, **5**, 130-143. https://doi.org/10.3923/ijcr.2009.130.143

[27] Idris Ali, A., Paul, V., Chattree, A., Prasad, R., Paul, A. and Amiteye, D. (2021) Evaluation of the Use of Different Solvents for Phytochemical Constituents and Antioxidants Activity of the Leaves of *Murraya koenigii* (Linn.) Spreng. (*Rutaceae*). *Plant Archives*, **21**, 985-992. https://doi.org/10.51470/plantarchives.2021.v21.no1.137

[28] Benzie, I.F. smf Strain, J.J. (1999) Ferric Reducing/Antioxidant Power Assay: Direct Measure of Total Antioxidant Activity of Biological Fluids and Modified Version for Simultaneous Measurement of Total Antioxidant Power and Ascorbic Acid Concentration. *Methods in Enzymology*, **299**, 15-27. https://doi.org/10.1016/S0076-6879(99)99005-5

[29] Sasidharan, I. and Menon, A.N. (2010) Effects of Temperature and Solvent on Antioxidant Properties of Curry Leaf (*Murraya koenigii* L.). *Journal of Food Science and Technology*, **48**, 366-370. https://doi.org/10.1007/s13197-010-0134-x

[30] Agarwal, R. (2013) Anticholinesterase, Antioxidant and Nitric Oxide Scavenging Activity of the Aqueous Extract of Some Medicinal Plants. *British Journal of Pharmaceutical Research*, **3**, 807-816. https://doi.org/10.9734/bjpr/2013/4833

[31] Apostolidis, E., Kwon, Y.I. and Shetty, K. (2007) Inhibitory Potential of Herb, Fruit, and Fungal-Enriched Cheese against Key Enzymes Linked to Type 2 Diabetes and Hypertension. *Innovative Food Science & Emerging Technologies*, **8**, 46-54. https://doi.org/10.1016/j.ifset.2006.06.001

[32] Mosmuller, E.W.J., Van Heemst, J.D.H., Van Delden, C.J., Franssen, M.C.R. and Engbersen, J.F.J. (1992) A New Spectrophotometric Method for the Detection of Lipase Activity Using 2, 4-Dinitrophenyl Butyrate as a Substrate. *Biocatalysis*, **5**, 279-287. https://doi.org/10.3109/10242429209014874

[33] Rice-Evans, C., Miller, N. and Paganga, G. (1997) Antioxidant Properties of Phenolic Compounds. *Trends in Plant Science*, **2**, 152-159. https://doi.org/10.1016/s1360-1385(97)01018-2

[34] Ningappa, M.B., Dinesha, R. and Srinivas, L. (2008) Antioxidant and Free Radical Scavenging Activities of Polyphenol-Enriched Curry Leaf (*Murraya koenigii* L.) Ex-

tracts. *Food Chemistry*, **106**, 720-728.
https://doi.org/10.1016/j.foodchem.2007.06.057

[35] Lee, J., Kim, H., Kim, J. and Jang, Y. (2002) Antioxidant Property of an Ethanol Extract of the Stem of *Opuntia ficus-Indica* Var. *saboten*. *Journal of Agricultural and Food Chemistry*, **50**, 6490-6496. https://doi.org/10.1021/jf020388c

[36] Singh, R.P., Chidambara Murthy, K.N. and Jayaprakasha, G.K. (2001) Studies on the Antioxidant Activity of Pomegranate (*Punica granatum*) Peel and Seed Extracts Using *in Vitro* Models. *Journal of Agricultural and Food Chemistry*, **50**, 81-86.
https://doi.org/10.1021/jf010865b

[37] Matsushige, K., Banset, P., Kadota, S. and Namba, T. (1996) Potent Free Radical Scavenging Activity of Dicaffeyl Quinic Acid Derivatives from Propolis. *Traditional Medicine*, **13**, 217-228.

[38] Gul, M.Z., Attuluri, V., Qureshi, I.A. and Ghazi, I.A. (2012) Antioxidant and *α*-Glucosidase Inhibitory Activities of *Murraya koenigii* Leaf Extracts. *Pharmacognosy Journal*, **4**, 65-72. https://doi.org/10.5530/pj.2012.32.12

[39] Kohen, R. and Nyska, A. (2002) Invited Review: Oxidation of Biological Systems: Oxidative Stress Phenomena, Antioxidants, Redox Reactions, and Methods for Their Quantification. *Toxicologic Pathology*, **30**, 620-650.
https://doi.org/10.1080/01926230290166724

[40] Lee, S., Lin, H. and Chen, C. (2008) Acylated Flavonol Monorhamnosides, A-Glucosidase Inhibitors, from Machilus Philippinensis. *Phytochemistry*, **69**, 2347-2353. https://doi.org/10.1016/j.phytochem.2008.06.006

[41] Sun, L., Gidley, M.J. and Warren, F.J. (2017) The Mechanism of Interactions between Tea Polyphenols and Porcine Pancreatic Alpha-amylase: Analysis by Inhibition Kinetics, Fluorescence Quenching, Differential Scanning Calorimetry and Isothermal Titration Calorimetry. *Molecular Nutrition & Food Research*, **61**, Article ID: 1700324. https://doi.org/10.1002/mnfr.201700324

[42] Palaniswamy, U.R. (2001) Human Dietetics and Asian Food Crops. *HortTechnology*, **11**, 504-509. https://doi.org/10.21273/horttech.11.4.504

[43] Indu Sasidharan, I.S. and Menon, A.N. (2010) A Study of Antioxidant Properties of Different Extracts of Curry Leaf (*Murraya koenigii* L.). *Electronic Journal of Environmental, Agricultural & Food Chemistry*, **9**, 1036-1046.
https://www.cabidigitallibrary.org/doi/pdf/10.5555/20103324693

[44] Wang, L., Yen, J.H., Liang, H.L. and Wu, M.J. (2020) Antioxidant Effect of Methanol Extracts from Lotus Plumule and Blossom (*Nelumbo nucifera* Gertn.). *Journal of Food and Drug Analysis*, **11**, Article 3. https://doi.org/10.38212/2224-6614.2726

[45] Shimada, K., Fujikawa, K., Yahara, K. and Nakamura, T. (1992) Antioxidative Properties of Xanthan on the Autoxidation of Soybean Oil in Cyclodextrin Emulsion. *Journal of Agricultural and Food Chemistry*, **40**, 945-948.
https://doi.org/10.1021/jf00018a005

[46] Tachibana, Y., Kikuzaki, H., Lajis, N.H. and Nakatani, N. (2001) Antioxidative Activity of Carbazoles from *Murraya koenigii* leaves. *Journal of Agricultural and Food Chemistry*, **49**, 5589-5594. https://doi.org/10.1021/jf010621r